INSTRUCTIONS



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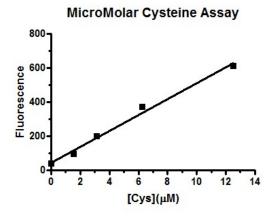
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ProFoldin MicroMolar Cysteine Assay Kit

CATALOG NUMBER CYS200

INTRODUCTION

Abnormal cysteine levels in vivo are related to cardiovascular disease, Huntington's disease, HIV infection and cancer. Cysteine is a common reducing agent in many in vitro experiments. The MicroMolar Cysteine Assay Kit is designed for measurement of micromolar concentrations of cysteine. The assay is based on increase of fluorescence at 535 nm of the dye R53 in the presence of cysteine. The assay kit can be used for measurements cysteine concentrations in biochemical reactions, pharmaceutical products and environmental water samples. The assay is compatible with HEPES buffer. It is not compatible with other thiol compounds such as DTT. For measurement of DTT, see the information of MicroMolar DTT Assay Kit (Catalog Number: DTT200).



The MicroMolar Cysteine Assay Kit (catalog number CYS200) includes 400 μ l of 100 x Dye R53. It is for measurement of 200 samples using 96-well plates. Cuvettes or 383-well plates may also be used for measurements.

Reference

Ho M.L et al, Silver nanoprism-based paper as ratiometric sensor for extending biothiol detection in serum. New Journal of Chemistry. Oct 30 (2017).

Wang LJ et al, Increased serum levels of cysteine in patients with schizophrenia: A potential marker of cognitive function preservation. Schizophrenia Research, Volume 192, Pages 391–397 (2018).

PROTOCOL

The following assay protocol is based on using a 96-well plate for the measurement. The sample volume is 100 μ l and the final assay volume is 300 μ l. For 384-well plate assays, the sample volume is 30 μ l and the final assay volume is 90 μ l. For assays using cuvette, the sample volume is 333 μ l and the final assay volume is 999 μ l.

INSTRUCTIONS



STANDARD CURVE

1. **Sample preparation:** Freshly prepare 100 μ l of cysteine solutions in a 96-well black plate with a two-fold serial dilution from 0.050 mM to zero in water or a 10 mM HEPES, pH 7.4 buffer. For 10 samples, dilute 22 μ l of the 100 x R53 dye 100-fold with water to make 2.2 ml of 1 x R53 dye.

2. **Detection:** Mix 200 μ l of 1 x dye R53 with 100 μ l of the cysteine solutions and immediately read the fluorescence at 535 nm with excitation at 485 nm.

Note: Cysteine is quickly oxidized by oxygen in the air or solution. The cysteine solution at low concentrations must be freshly prepared before the measurement. After mixing the dye and the cysteine sample, the fluorescence intensity is read immediately.

3. Data Analysis: Plot the fluorescence intensity Fc and the cysteine concentration [Cysteine] to generate the linear standard curve.

Fc = a [Cysteine] + b

Where the **Fc** values are from experimental data, the **a** and **b** values are from the linear fitting between the **Fc** values and the cysteine concentrations.

UNKNOWN SAMPLES

Follow the same procedure to measure the fluorescence intensity Fc values from the unknown samples. Calculate the cysteine concentrations in the unknown samples using the Fc values from the unknown samples and the **a** and **b** values from the standard curve.

[Cysteine] = (Fc - b) / a

RELATED PRODUCTS

MicroMolar Histidine Assay Kit	Catalog number: HIS200
MicroMolar Primary Amine Assay Kit	Catalog number: PAA100K
MicroMolar UDP Assay Kit	Catalog number: MUD100K
MicroMolar EDTA Assay Kit	Catalog number: EDTA200
MicroMolar DTT Assay Kit	Catalog number: DTT200
NanoMolar Zinc Assay Kit	Catalog number: NZA1000
MicroGram Lipid Assay Kit	Catalog number: LIP1000

For more concentration assays of various biochemical molecules and inorganic ions, please visit our website at www.profoldin.com.